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## **Agricultural diversification promotes multiple ecosystem services without compromising yield**

Tamburini, Giovanni ; Bommarco, Riccardo ; Wanger, Thomas Cherico ; Kremen, Claire ; van der Heijden, Marcel G A ; Liebman, Matt ; Hallin, Sara

**Abstract:** Enhancing biodiversity in cropping systems is suggested to promote ecosystem services, thereby reducing dependency on agronomic inputs while maintaining high crop yields. We assess the impact of several diversification practices in cropping systems on above- and belowground biodiversity and ecosystem services by reviewing 98 meta-analyses and performing a second-order meta-analysis based on 5160 original studies comprising 41,946 comparisons between diversified and simplified practices. Overall, diversification enhances biodiversity, pollination, pest control, nutrient cycling, soil fertility, and water regulation without compromising crop yields. Practices targeting aboveground biodiversity boosted pest control and water regulation, while those targeting belowground biodiversity enhanced nutrient cycling, soil fertility, and water regulation. Most often, diversification practices resulted in win-win support of services and crop yields. Variability in responses and occurrence of trade-offs highlight the context dependency of outcomes. Widespread adoption of diversification practices shows promise to contribute to biodiversity conservation and food security from local to global scales.

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## ECOLOGY

# Agricultural diversification promotes multiple ecosystem services without compromising yield

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Enhancing biodiversity in cropping systems is suggested to promote ecosystem services, thereby reducing dependency on agronomic inputs while maintaining high crop yields. We assess the impact of several diversification practices in cropping systems on above- and belowground biodiversity and ecosystem services by reviewing 98 meta-analyses and performing a second-order meta-analysis based on 5160 original studies comprising 41,946 comparisons between diversified and simplified practices. Overall, diversification enhances biodiversity, pollination, pest control, nutrient cycling, soil fertility, and water regulation without compromising crop yields. Practices targeting aboveground biodiversity boosted pest control and water regulation, while those targeting belowground biodiversity enhanced nutrient cycling, soil fertility, and water regulation. Most often, diversification practices resulted in win-win support of services and crop yields. Variability in responses and occurrence of trade-offs highlight the context dependency of outcomes. Widespread adoption of diversification practices shows promise to contribute to biodiversity conservation and food security from local to global scales.

## INTRODUCTION

Agricultural expansion and intensification are considered major drivers of habitat and biodiversity loss, soil and freshwater degradation, environmental pollution, and greenhouse gas emissions worldwide (1, 2). Implementation of a new crop production paradigm is needed to take on the local to global challenges of providing food security for rapidly growing demands from human societies while minimizing negative impacts on the environment in a world exposed to global changes (3).

Crop management based on diversification practices that enhance key elements of biodiversity has been suggested to reduce impacts on the environment without negative effects on crop yields (4). Enhancing the diversity of biological communities, both above and below ground, can increase resource use efficiency and the stability of ecosystem production over time (5–7). Agricultural diversification is the intentional addition of functional biodiversity to cropping systems at multiple spatial and/or temporal scales, and it aims at regenerating biotic interactions underpinning yield-supporting ecosystem services (8). It embraces a variety of practices encompassing the management of crops, noncrop habitats, soil, and landscapes (9). Functional biodiversity can be enhanced by increasing crop species diversity (e.g., intercropping and crop rotation), increasing noncrop species diversity within and around the fields (e.g., flower strips, hedgerows, and seminatural habitats), or by in-

oculation of beneficial microorganisms into the soil (e.g., arbuscular mycorrhizal fungi, N<sub>2</sub>-fixing bacteria, and growth-promoting bacteria). Functional diversity below ground can also be supported and stimulated through addition of organic inputs (e.g., manure and crop residues) or reducing soil disturbance (e.g., reduced tillage), which lead to soil stratification and thereby more niches (8, 9). Despite a rapidly growing body of research assessing the impacts of agricultural diversification practices on biodiversity and related ecosystem services (7, 10–12), there is no comprehensive quantitative synthesis of this information. Consequently, we lack the broader understanding of whether diversification practices are actually capable of supporting biodiversity and multiple ecosystem services, including crop yields.

We investigated the impact of multiple agricultural diversification practices on biodiversity and related ecosystem services and compared these with cropping systems with less diverse farming practices typical of mainstream agriculture. First, we systematically reviewed published meta-analyses and summarized the number of reported effect sizes (vote count) to assess the current state of knowledge, identify research gaps, and explore general patterns. We included studies based on stringent criteria such as relevance, eligibility, and statistical independence, thereby ensuring the largest possible primary database with minimum overlap of original studies (figs. S1 and S2 and see Materials and Methods). Second, to estimate the overall impact of agricultural diversification on biodiversity and ecosystem services provisioning, we performed a second-order meta-analysis on the subset (70%) of the meta-analyses that reported comparable effect sizes. Second-order meta-analyses are frequently used in health science but have only rarely been used in ecology or agricultural research (see Materials and Methods).

We based our systematic review on 98 meta-analyses and 456 effect sizes based on 6167 original studies (see Materials and Methods). We grouped the diversification practices into six broad categories [following (8, 9)]. The first five are crop diversification by addition of crop species in the field over space or time, noncrop diversification by addition of noncrop habitats within or around the field or in the surrounding landscape, organic amendment by addition of organic

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material to the soil, inoculation by addition of beneficial microorganisms into the soil, and reduced tillage (table S1). In addition, we included organic farming, i.e., production systems free of synthetic pesticides and mineral fertilizers, as a sixth separate category for comparison since this is a widely adopted practice in some regions. In the selected meta-analyses, diversified agriculture was compared with the relevant control practices typical of mainstream farming, such as monoculture, short crop rotation, simplified landscapes, use of mineral fertilizers, and deep tillage (table S1). We divided the response variables into nine biodiversity/ecosystem service categories (hereafter, “ecosystem service categories”) (13, 14): biodiversity, water regulation, carbon sequestration, climate regulation, nutrient cycling, soil fertility, pollination, pest control, and crop yield (table S2). Water regulation refers to water quality and quantity, climate regulation refers to greenhouse gas dynamics, and carbon sequestration refers to carbon storage. To describe the effects of diversification practices on key attributes of cropping systems comprehensively, we kept soil fertility, nutrient cycling, carbon sequestration, and climate regulation as separate services, although the first two have been previously grouped as “maintenance of soil fertility” (14) and the last two as “climate regulation” (13). In our systematic review, for each ecosystem service category, we recorded and compared the number of different responses to diversification (i.e., effect sizes), classifying them as positive, negative, or neutral (i.e., no significant effect).

For the second-order meta-analysis, we selected 69 meta-analyses, enabling a statistical analysis of 324 effect sizes, based on 5160 original studies and a total of 41,946 original comparisons. We extracted global effect sizes, sampling error variances, and their associated sample sizes, i.e., the number of original comparisons. We transformed effect sizes to a common metric [log of the response ratio (lnRR)] and conducted multilevel mixed-effects meta-analyses to explore effects of diversification practices on biodiversity and ecosystem service delivery (see Materials and Methods). We found an unbalanced occurrence of effect sizes belonging to different ecosystem service categories across diversification practices and, therefore, adopted a multiple step hierarchical approach. We first ran a global model to test whether the mean effect of diversification differed from zero for different ecosystem service categories with all diversification practices included (model 1) (see Materials and Methods). Second, we explored biodiversity and ecosystem service responses to diversification practices mainly targeting functional biodiversity in the aboveground environment with crop and noncrop diversification (model 2) and the belowground environment with organic amendment, reduced tillage, and inoculation (model 3). Third, we ran separate models for practices that had a sufficient number of effect sizes, which were organic amendment, reduced tillage, and crop diversification (models 4, 5, and 6, respectively). All models included sample size as a weight, thus giving more importance to effect sizes based on a higher number of original studies and comparisons. The results from both the systematic review and the second-order meta-analyses were robust to variations in study quality, inclusion criteria, and ecosystem service classification and to potential publication bias (figs. S3 to S6).

## RESULTS

### Trends from the systematic review

Our systematic review showed that the impact of agricultural diversification on biodiversity and ecosystem services was predominantly

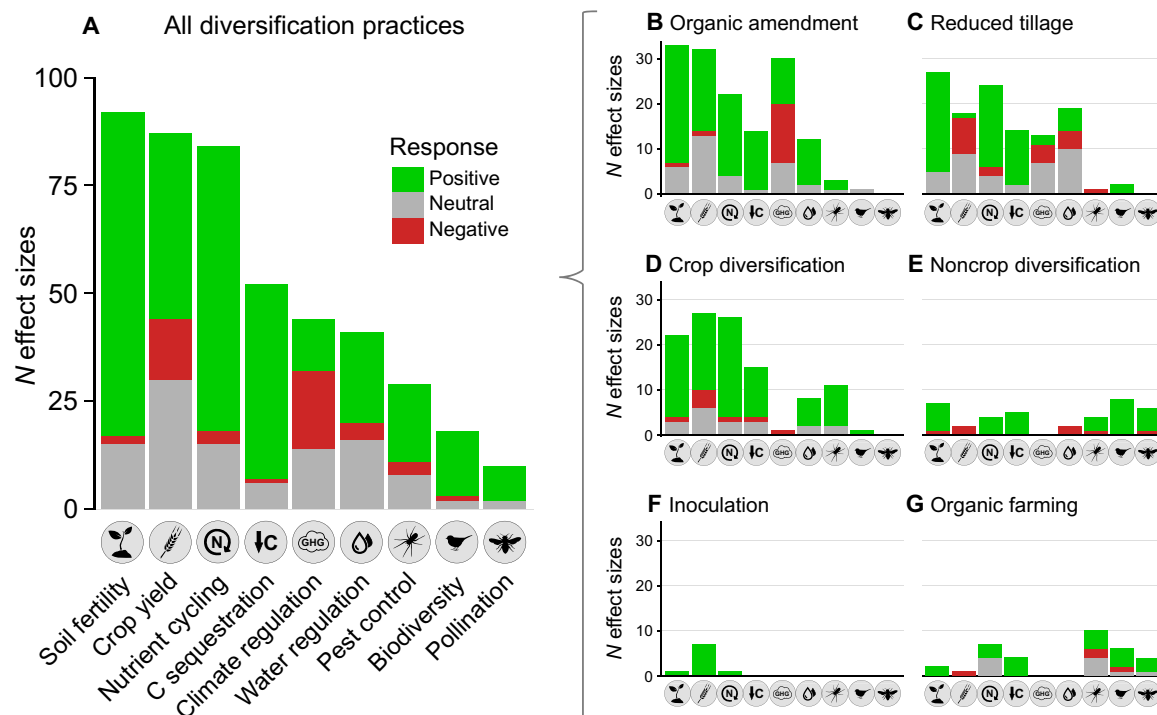
positive (67% positive effect sizes, 23% neutral effect sizes, and 10% negative effect sizes; Fig. 1), with soil fertility and nutrient cycling having the highest numbers of positive effect sizes. Crop yield and climate regulation displayed the highest proportion of negative responses to diversification practices (16 and 41%, respectively; Fig. 1A). The number of published meta-analyses has increased exponentially in the past decade, especially global-scale analyses (fig. S7, A and C). However, we found research gaps for specific services and practices. The most frequently observed ecosystem services category was soil fertility with 92 effect sizes, followed by crop production and nutrient cycling (87 and 84 effect sizes; fig. S7D). By contrast, pollination, biodiversity, and pest control were less represented (10, 18, and 29 effect sizes, respectively). The most examined diversification practices were organic amendment, reduced tillage, and crop diversification (146, 118, and 111 effect sizes), whereas noncrop diversification and inoculation, as well as organic farming, were less represented (38, 9, and 34 effect sizes) and need further investigations.

### Biodiversity and ecosystem service response to diversification

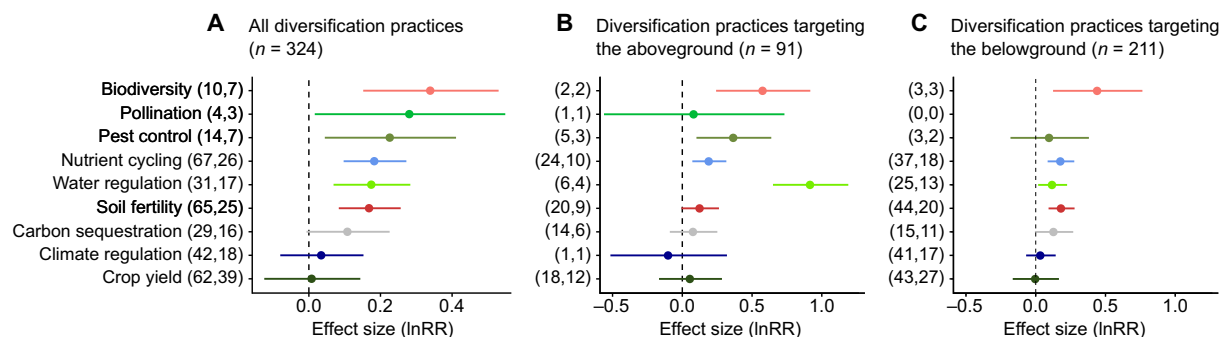
The second-order meta-analysis showed that agricultural diversification strengthens several ecosystem service categories (omnibus test  $Q_M = 43.67$ ;  $P < 0.0001$ ; Fig. 2A) while having a neutral effect on crop yield [lnRR = 0.01; 95% confidence interval (CI) = -0.12 to 0.14]. Diversification practices enhanced biodiversity (lnRR = 0.34; 95% CI = 0.15 to 0.53), pollination (lnRR = 0.28; 95% CI = 0.02 to 0.55), pest control (lnRR = 0.23; 95% CI = 0.04 to 0.41), nutrient cycling (lnRR = 0.18; 95% CI = 0.10 to 0.27), water regulation (lnRR = 0.18; 95% CI = 0.07 to 0.28), soil fertility (lnRR = 0.17; 95% CI = 0.08 to 0.26) and, marginally, carbon sequestration (lnRR = 0.11; 95% CI = -0.01 to 0.23). Climate regulation response did not differ statistically from a neutral effect (lnRR = 0.04; 95% CI = -0.08 to 0.15). Both groups of diversification practices targeting either the above- or belowground environment enhanced nutrient cycling and water regulation, but affected the delivery of other services differently (Fig. 2, B and C, and table S4). Diversification practices targeting the aboveground environment (i.e., crop and noncrop diversity) increased pest control, whereas diversification practices targeting the belowground environment (i.e., organic amendment, reduced tillage, and inoculation) enhanced soil fertility and, marginally, carbon sequestration (Fig. 2, B and C, and table S4). Analyses of specific practices presented consistent results: organic amendment increased water regulation, soil fertility, nutrient cycling, and carbon sequestration, reduced tillage enhanced soil fertility, nutrient cycling, and (belowground) biodiversity, and crop diversity improved (aboveground) biodiversity, pest control, nutrient cycling, and water regulation (table S5).

### Trade-offs between crop yield and multiple ecosystem services

We visualized trade-offs, lose-lose relationships, and win-win relationships between crop yield and multiple services by plotting combinations of effect sizes (15) gathered from the 23 meta-analyses in which the responses to diversification of crop yield and at least one other service were analyzed simultaneously (Fig. 3). We only found 111 effect size combinations, highlighting that effects of diversification on multiple ecosystem services is a major research gap (16). We found that agricultural diversification mainly promoted win-win



**Fig. 1. Vote count reveals that agricultural diversification practices generally have a positive impact on biodiversity and ecosystem services.** Number of reported effect sizes with a significant positive (green), negative (red), or neutral (gray) response to agricultural diversification, overall (A) and to each category of diversification practice separately (B to G). The systematic review comprises 456 effect sizes from 98 meta-analyses based on 6167 original studies (fig. S1). Diversification practice and ecosystem service categories were based on classifications following (8,9) and (13, 14, 27), respectively (tables S1 and S2).

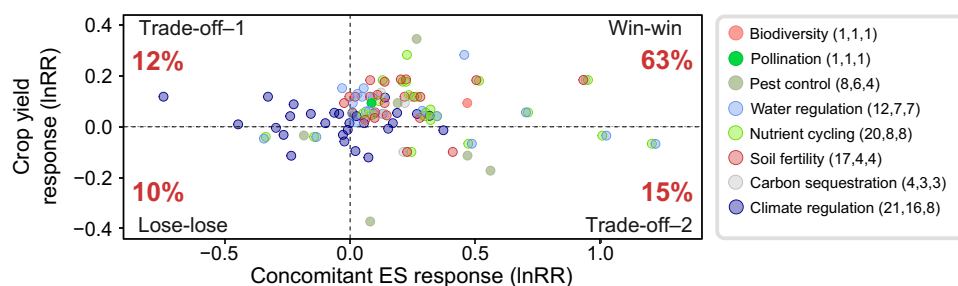


**Fig. 2. Second-order meta-analysis shows how agriculture diversification promotes biodiversity and ecosystem services without compromising crop yield when compared with cropping systems without these practices.** (A) All diversification practices included (324 effect sizes and 69 meta-analyses, based on 5160 original studies with 41,946 comparisons). (B) Diversification practices targeting the aboveground environment (crop and noncrop diversity; 91 effect sizes and 24 meta-analyses). (C) Diversification practices targeting the belowground environment (organic amendment, reduced tillage, and inoculation; 211 effect sizes and 55 meta-analyses). Note the difference in scale of the x axes when comparing (A) with (B) and (C). Organic farming is included only in the global model (A) since it often includes practices targeting both above- and belowground environments. The number of effect sizes and meta-analyses included in each category are displayed in parentheses. Ecosystem service categories are classified following (13, 14, 27) (table S2). Error bars represent 95% CIs.

scenarios, supporting crop yield and the provisioning of a range of services (63% of the combinations; Fig. 3). Most of them are moderate gains, but the greatest win-win relationships with crop yield include nutrient cycling and soil fertility. Climate regulation presented instead the highest number of trade-offs, with 50% representing situations where an increase in yield corresponded to a decrease in climate regulation provision. In addition, pest control was competing with yield in some cases.

## DISCUSSION

Both the systematic review and the second-order meta-analysis show that agricultural diversification practices enhance biodiversity and the delivery of several supporting and regulating ecosystem services pivotal to crop yield. Crop and noncrop diversification increased the provisioning of pest control and pollination, respectively (Figs. 1, D and E, and 2), which is in line with global results based on raw primary data (17). Services associated with soil quality, particularly



**Fig. 3. Agricultural diversification generally promotes win-win scenarios, simultaneously supporting crop yield and the provisioning of a concomitant ecosystem service category.** The visualization is based on a subset of meta-analyses, which simultaneously presented the responses to agricultural diversification of crop yield (y axis) and at least one concomitant ecosystem service (ES) (x axis) (in total 24 studies, 111 pairs of effect sizes). Numbers in red indicate the proportion of effect size combinations in each quadrante. Points represent combinations of raw effect sizes (lnRR) and the colors correspond to the specific service, as indicated in the box to the right. Values in parentheses after each service indicate the number of effect sizes for the concomitant service, crop yield, and the number of meta-analyses.

soil fertility and nutrient cycling, responded in a consistent positive manner to several diversification practices, mainly to organic amendment and reduced tillage (Fig. 1), and presenting the smallest variabilities (Figs. 2 and 3). This is likely because these services are largely affected by the soil organic carbon pool, which is typically promoted by diversification measures (2). Our sensitivity analysis revealed that the similar responses of soil services were not merely due to using common indicators or correlations among variables (e.g., soil organic carbon; table S2), but rather similar responses of different soil functions and properties (figs. S4 and S5). Diversification practices also increased both the quality and quantity components of water regulation provisioning (fig. S6). In particular, practices targeting the aboveground environment, i.e., crop and noncrop diversity, greatly enhanced water regulation (Fig. 2B), primarily by increasing water quality by limiting nitrogen leaching loss (table S2). Differences in effect sizes when separating between above- and below-ground diversification practices suggest different mechanisms and processes driving this ecosystem service response to diversification.

The variation in crop yield response to overall diversification suggests a high degree of context dependency (Figs. 1 and 2). It highlights that there are conditions and practices we need to avoid but also ample opportunities to reap benefits from diversification. For example, our systematic review shows that yield often decreases under reduced tillage management and organic farming but generally increases with crop diversification and inoculation (Fig. 1, C, D, F, and G). Moreover, yields have been shown to improve under reduced tillage in dry climates by retaining crop residues and by applying longer crop rotations (18). The adoption of appropriate combinations of diversification practices thus holds great potential to increase yields compared with mainstream farming. The visualization of the relationships between crop yield and multiple services reveals that the majority of the win-win situations (74%) includes soil fertility, nutrient cycling, and water regulation (Fig. 3), probably because practices that improve soil functioning simultaneously increase resource availability for the crops. A key aim for the development of locally adapted cropping systems will be to identify diversification solutions that sustain both crop yield and multiple ecosystem functions, resulting in win-win outcomes.

The response of climate regulation was highly variable and yielded the highest number of trade-offs and lose-lose relationships (Fig. 3). These were mostly driven by increased greenhouse gas emissions caused by the application of organic soil amendments (Fig. 1)

(3, 19). However, our analysis also demonstrates that organic inputs promote soil fertility, nutrient cycling, carbon sequestration, and water regulation, by increasing soil organic carbon, nutrient availability, and soil water storage and limiting nutrient leaching and runoff (tables S2 and S5). Soil bacteria and arbuscular mycorrhizal fungi, which are key contributors to soil functioning, were also enhanced (fig. S6 and table S2) (6). Organic amendments thus have complex positive and negative and also cascading effects on the cropping system and the environment. For example, positive climate feedbacks can arise because of increased soil respiration caused by priming effects, where added organic material triggers the degradation of older soil organic matter, leading to emissions of CO<sub>2</sub>, but are mainly driven by the increasing emissions of the far more potent greenhouse gas nitrous oxide. Organic amendments fuel microorganisms with the capacity to produce this gas to such a degree that the positive climatic effects by increased carbon storage in fertilized agricultural soils are predicted to be offset by nitrous oxide emissions already by 2060 (20). It would be valuable to further consider not only the impacts of organic matter input but also those of production, such as livestock and cultivation of legumes for feed with large impact on land-use change and direct greenhouse gas emissions. This would allow a comprehensive understanding for effects of organic amendments on climate change. A key challenge for sustainable crop production is hence to seek diversification solutions that simultaneously sustain soil health, crop yield, and mitigation of climate change.

We show that agricultural diversification promotes biodiversity and the delivery of ecosystem services without compromising crop yield. This suggests several pathways for future sustainable food production and demonstrates how mainstream, high-yielding crop production can benefit from management that bolsters biodiversity. Large areas of cropland with monocultures in vast crop fields, such as in Australia, and North and South America (3), could benefit by the implementation of longer crop rotations, intercropping, and higher noncrop species diversity that enhance aboveground biodiversity and the provisioning of regulating services, such as pollination, pest control, and water regulation. Belowground biodiversity and services associated with soil quality are mainly supported by organic amendment and reduced tillage. Further, our results indicate that combining diversification practices is potentially advantageous for the provisioning of multiple ecosystem services and biodiversity conservation (Fig. 2). This shows promise to reduce crop production



dependency on agrochemicals and its negative impacts on the environment, adapt to and hedge risks from climate change, and contribute to global food security.

Overall, agricultural diversification emerges as a general strategy to reach the sustainable development goals defined by the United Nations, which basically all are directly or indirectly linked to agriculture. Trans- and interdisciplinary research efforts will be required to tailor economically, socially, and environmentally sustainable diversification practices to specific cropping systems and local contexts. Although recent research shows that diversification can be an economically viable alternative for farmers also in current food systems (21), widespread uptake of diversified agriculture needs to be supported by transformations in the food systems. This involves investments into the development and spread of evidence-based knowledge on the efficiency of diversification practices, including cost-benefit analyses (22), and into the removal of potential barriers to farmers' uptake such as up-front costs, access to credit and appropriate equipment, as well as into supportive technologies and infrastructure to process and distribute the products. Changes of market conditions causing imbalanced power relations in the food value chain (23) and improved targeting of subsidies are needed to relieve farmers from the price-cost squeeze of high input and low output prices (24). This would provide farmers with the decision space, tools, and opportunity to develop and implement diversification practices based on situated knowledge.

## MATERIALS AND METHODS

### Systematic review

We focused on meta-analyses rather than on original studies because they are useful for summarizing scientific evidence and extrapolating general trends (25). The main advantage is the high level of generalization that can be achieved when using a large number of individual observations already summarized in first-order meta-analyses. Moreover, since a meta-analysis can be conducted only when sufficient and appropriate quantitative data from original studies are available, the number of published meta-analyses about a certain topic is an indicator of the overall understanding of the subject, allowing us to reveal research gaps (25, 26).

### Definition of diversification farming practices and ecosystem services

Diversification practices were defined as intentional addition of functional biodiversity to cropping systems at multiple spatial and/or temporal scales (8). The six categories of diversification practices were based on previous classifications and selected in an expert workshop to cover the diversification practices found in our systematic review (table S1). We acknowledge that other classifications are possible and some management categories overlap. For instance, organic farming often also includes other diversification practices as crop rotation and the application of organic amendments.

Numerous concepts and classification systems for ecosystem services have emerged in the past decades leading to a plurality in terminology and definitions. We defined ecosystem services as “the direct and indirect contributions of ecosystems to human well-being” (14). Building on existing classifications (13, 14, 27), we identified eight ecosystem services in the cropping system context: water regulation, carbon sequestration, climate regulation, nutrient cycling, soil fertility, pollination, pest control, and crop yield (table S2). The

first seven services affect crop yield directly in the field and also indirectly by influencing the larger environment around the field. Some of these services also affect human well-being via other mechanisms than their contribution to food production, e.g., carbon sequestration and clean water.

### Literature search and selection criteria

A flowchart of the literature search and selection and study framework is provided in fig. S1 (28). We initiated a literature search using the Web of Science online database using the search string:

TOPIC = (crop\* OR “noncrop” OR intercropping OR “intercropping” OR “inter cropping” OR compost\* OR till\* OR “vegetation strip\*” OR agroforest\* OR inoculat\* OR landscape OR organic OR fallow OR conventional OR fert\* OR reduce\* OR rotation OR “catch crop” OR amend\*) AND (“meta-analysis” OR “metaanalysis” OR “meta-analysis”) AND (divers\* OR soil\* OR biomass\* OR water\* OR pollutant\* OR sediment\* OR fodder OR emission\* OR carbon\* OR climat\* OR pest\* OR “biocontrol” OR weed\* OR pollinat\* OR fert\* OR energ\* OR resistance OR productiv\* OR yield\*). The search was then refined for the categories Environmental Sciences, Environmental Studies, Ecology, Agronomy, Agriculture Multidisciplinary, Plant Sciences, Biodiversity Conservation, Evolutionary Biology, Horticulture, Multidisciplinary Sciences, Entomology, Biology, and Soil Sciences. We identified peer reviewed meta-analyses published until November 2018. We then integrated the literature search with targeted research strings in Google Scholar (e.g., “meta analysis AND crop AND “flower strips” OR “hedgerow”). We achieved a reasonable coverage of grey literature since many of the meta-analyses were partly based on unpublished data and non-English texts not indexed in official search engines.

We examined the title and abstract of each article to assess how well it met our selection criteria. The main selection criterion required a quantitative assessment of the impact of one or more diversification practices on any ecological process in the context of crop production (table S1). This resulted in an initial set of 278 articles, for which the following additional criteria for inclusion were adopted: (i) The effect of a diversification practice had to be investigated for crop species, including biofuel crops, and compared with that of a control, i.e., farming practices commonly adopted in mainstream agriculture and standard for that particular crop and region. We excluded studies on livestock production systems, comparisons of cropping systems with natural or seminatural ecosystems, and transitions from crop fields to other habitat types, i.e., afforestation or crop land abandonment. (ii) The list of original studies included in the meta-analyses and the origin of grey literature had to be presented. In five cases, we asked authors to provide the reference list. We excluded any meta-analyses whose original studies were a subset of a more recent meta-analysis. (iii) The effect of a diversification practice on the response variables had to be calculated with meta-analytic techniques and presented as an effect size. We excluded studies that mentioned an effect size without specifying how it was calculated. (iv) We only included the global effects if partial effect sizes based on subsets of observations were also presented, to avoid pseudo-replication. For the same reason, if potentially correlated response variables were investigated, then we made an informed decision on which variable to select. For instance, if both soil organic carbon and total soil carbon were presented in the study, then we chose only soil organic carbon. (v) For multiple effect sizes calculated for different points across space, e.g., at different soil depths, we

included only the measures most relevant to crop production, such as soil organic matter measured between 0- and 30-cm soil depth. A total of 111 meta-analyses met our criteria.

### Statistical independence assessment

Extrapolating general trends from a set of meta-analyses requires the fundamental assumption that the primary meta-analyses are independent, i.e., that they are based on different sets of original studies. However, it is possible that an original study is included in more than one meta-analysis on a given topic. This happens when different meta-analyses ask different questions or when data from older meta-analyses have been included in more recent analyses. To avoid this form of pseudo-replication, we examined more than 6300 references (fig. S2) and calculated the proportion of shared original studies among the selected meta-analyses. We included only meta-analyses presenting a maximum of 30% of shared original studies (29). We checked for both the systematic review and the second-order meta-analysis whether the adoption of more stringent thresholds affected our results (see “Sensitivity analysis” section below). If two or more meta-analyses shared a higher proportion than 30%, then we selected the meta-analysis based on the largest number of original studies (usually the most recent) and discarded the others (i.e., last, none had a count of overlapping studies beyond 30%). Meta-analyses presenting shared original studies, but focusing on different response variables, e.g., effect of tillage management on soil organic carbon versus effect on yield, were not considered as a source of pseudo-replication.

### Data extraction and response variable assignment to different ecosystem service categories

A total of 98 meta-analyses met our criteria and were considered statistically independent. Of the selected meta-analyses, 80% had <20% shared original studies (mean value for the entire dataset, 11.5%). For each article, we further extracted the geographical range covered by the original studies, the type of diversification practice investigated, the response variables measured, and the number of original comparisons for each effect size (fig. S7). We also collected additional details for each effect size (where present) about specific methodologies (data file S1). We categorized the response variables of the included meta-analyses in different types of ecosystem services or biodiversity (table S2). Assignment to categories was performed by the coauthors according to their expertise in soil sciences (S.H., M.G.A.v.d.H., and M.L.), biodiversity (S.H., C.K., M.G.A.v.d.H., R.B., T.C.W., and G.T.), pollination and pest control (C.K., R.B., T.C.W., M.L., and G.T.), and microbiology (S.H. and M.G.A.v.d.H.). While some response variables were direct measures of the final service delivered (e.g., crop yield), others represented the ecosystem functions underpinning those services (e.g., soil organic carbon content for soil fertility, and predation for pest control) or were general indicators (informative proxies) (30) of the service (e.g., weed and pollinator abundance for weed control and pollination, respectively). Some of the response variables were positively related to service provisioning, whereas others were negatively related. For the systematic review, we recorded whether the effect of a given diversification practice on a particular response variable was significantly positive, neutral, or significantly negative. We then adjusted the direction considering the relationship between each response variable and the correlated service provision. For example, a significant decrease in carbon dioxide emissions in

response to biochar input was reported as a significant positive effect of organic amendment on climate regulation.

Many response variables affect or are proxies for multiple categories of ecosystem services. For example, pollinator diversity can be considered as a measure of both pollination and biodiversity. In these cases, we used the same effect size for different ecosystem service categories. The most frequent instance involved measures of soil organic matter content categorized as measures of soil fertility, nutrient cycling, and carbon sequestration (table S2) (31). We checked for both the systematic review and the second-order meta-analysis whether the inclusion of repeated effect sizes in the dataset affected our results (see the “Sensitivity analysis” section below). The final dataset comprised 456 effect sizes of which 113 were repeated, from 98 meta-analyses based on 6167 original studies.

### Quality assessment of individual meta-analyses

For each selected meta-analysis, we calculated a methodological quality index since poorly and inexpertly conducted meta-analyses can provide biased and misleading results (24, 32). We used eight criteria to assess study quality based on availability and/or clarity in the presentation of (i) definition of the control group, (ii) literature search method, (iii) number of original studies, (iv) inclusion/exclusion criteria of original studies, (v) effect size average and CIs, (vi) weighting procedure, (vii) heterogeneity assessment, and (viii) sensitivity control. For each of these eight criteria, a meta-analysis could receive a score of either 1 (low quality) or 2 (high quality). We used the sum of the scores as an overall measure of quality, ranging from 8 to 13 (“low quality”) to 14 to 16 (“high quality”). Our dataset presented a score ranging from 10 to 16 (14 on average). We checked for both the systematic review and the second-order meta-analysis whether the inclusion of low-quality studies in the dataset affected our results (33) (see the “Sensitivity analysis” section below).

### Second-order meta-analysis

For the second-order meta-analysis, we first selected a subset of meta-analyses that reported comparable measures of effect sizes, such as the  $\ln RR$  or closely related metrics (e.g.,  $RR$  and percentage of change), where  $\ln RR = \ln(XE) - \ln(XC)$  ( $XE$ , diversification practice;  $XC$ , mainstream practice). This is the ratio of the outcome of an experimental group ( $XE$ ) to that of a control group ( $XC$ ). We excluded studies reporting metrics based on standardized mean differences (i.e., Hedges  $d$ ) or correlation coefficients because they cannot be transformed into an  $RR$  without access to the original data (34). On the basis of these criteria, we were able to include 69 meta-analyses. From each of these, we extracted global effect sizes, sampling error variances, and their associated sample sizes. Sampling error variance is the square of the standard error ( $SE$ ), but these estimates are rarely reported. Instead, a 95% CI of the effect size is usually provided, and half the width of the 95% CI divided by 1.96 is a good approximation to the  $SE$  (35, 36). Data were extracted from tables or graphs using GetData Graph Digitizer 2.26 (<http://getdata-graph-digitizer.com/>). We transformed effect sizes of individual meta-analyses to a common metric ( $\ln RR$ ). Since direct log transformation of an  $RR$  (19 effect sizes) can lead to an overestimation of the effect size, we applied a correction for the log transformation as  $\ln(RR) = \ln(RR) - NSE \times (2 RR2) - 1$ , where  $N$  is the number of original comparisons. When higher values of the effect sizes would mean negative impacts on service provisioning (e.g., abundance of pest species or  $CO_2$  emission), we reversed

the sign of the response. The final dataset for the second-order meta-analysis included 324 effect sizes from 69 meta-analyses based on 5160 original studies and 41,946 original comparisons.

### Data analysis

We first ran a multilevel mixed-effects meta-analysis to determine whether mean effect sizes for different ecosystem service categories differed from 0 (model 1). We performed the analyses in the metafor package (ver. 2.1, rma.mv function) (36) that incorporates both fixed (moderators) and random effects, allowing us to control for nonindependence in the data due to multiple effect sizes per meta-analysis. To account for heterogeneity both between and within studies, we specified the effect size ID (identifier) and the study ID as random effects in our model. The model that included a nested random structure (1|Study ID/Effect size ID) yielded the lowest Akaike Information Criterion (AIC) score compared with the other candidate structures (table S3), and it was therefore retained. We included the number of original comparisons as weight in the model (24, 35), giving more importance to measures based on a higher number of original studies and comparisons. Last, we included the different types of biodiversity/ecosystem services as one categorical moderator with nine levels in the model. The general form of the global model was

$$\ln RR \sim ES, v_i, \text{weight} = N, \text{random} = \sim 1 | \text{Study ID/Effect size ID}$$

where ES is the ecosystem service categories,  $v_i$  is sampling error variance, and  $N$  is the number of original comparisons. Including ES as moderator in the model led to a lower AIC score compared with the null model ( $\Delta AIC = 34.17$ ). The model was run without the intercept to obtain the parameter estimates (mean effect sizes) for each level of the categorical variable. We did not include the interaction between ES and different types of diversification practices because the number of combinations was too high (38 combination levels). To further investigate the effects of different diversification practices on biodiversity and ecosystem service delivery, we analyzed different subsets of data. We ran two models only considering diversification practices targeting above- or belowground environment (model 2: crop and noncrop diversification; model 3: organic amendment, reduced tillage, and inoculation). Organic management was not considered because it often includes both practices targeting above- and belowground environment (e.g., crop rotation and organic amendments). We then ran separate models for those practices with sufficient number of effect sizes, when considering the number of meta-analyses, the number of effect sizes and their distribution across explanatory variable's levels. We hence separately analyzed organic amendment (model 4, 126 effect sizes), reduced tillage (model 5, 82 effect sizes), and crop diversity (model 6, 72 effect sizes). Models 2, 5, and 6 included only the effect size ID as random effect, yielding the lowest AIC compared with the other candidate structures. We did not include the geographical range covered by the original studies as an explanatory variable in the analyses due to the unbalanced distribution of effect sizes across geographical regions, ecosystem service categories, and diversification practices. Most of the meta-analyses performed global analyses, and specific geographical areas were represented by only a few studies. Fixed factor estimates were considered statistically significant if the 95% CI did not overlap zero. We checked the profile likelihood plots to ensure the identifiability of the variance components in all the models (e.g., model 1; fig. S8) (36). All parameter estimates are

reported for the best model run with REML (restricted maximum likelihood). All analyses were performed in R software.

### Publication bias

Publication bias was assessed using Funnel plot and Egger test techniques by using meta-analytic residuals and including in rma.mv function the precision ( $1/SE$ ) as a covariate (33, 37). We considered analyses to be biased if the intercept of this regression significantly deviated from zero, indicating that the overall relationship between the precision and residuals is asymmetrical. We then calculated Rosenberg's fail-safe number to assess the robustness of our results to potential publication bias (38). Furthermore, we explored whether our results were driven by influential outliers, defined as effect sizes with hat values (i.e., diagonal elements of the hat matrix) greater than two times the average hat value (i.e., influential) and standardized residual values exceeding 3.0 (i.e., outliers) (39, 40). Funnel plot (fig. S3A), Egger test [intercept, 0.01 (95% CI:  $-0.07, 0.09$ ,  $P = 0.8502$ )], and the Rosenberg's fail-safe number (1131528,  $P < 0.0001$ ) showed no sign of publication bias. The graphical test for influential outliers was also negative (fig. S3B).

### Sensitivity analysis

We explored the robustness of the patterns from the systematic review and the results of the second-order meta-analysis to low-quality data, repeated effect sizes, and different thresholds of shared original studies among meta-analyses. We tested four datasets: (i) the complete dataset and three subsets including (ii) only high-quality studies, (iii) no repeated effect sizes, and (iv) only high-quality studies and no repeated effect sizes. For each of these, we applied four different maximum levels of shared original studies (30, 25, 20, and 15%). For the systematic review, results were highly robust as reflected in the similar responses patterns for the data subsets (fig. S4). For the second-order meta-analysis, we reran the analysis for each of the four datasets at different maximum levels of shared original studies. Again, results were robust as shown by similar effect sizes for the data subsets (fig. S5).

Given the heterogeneity of response variables included in each ecosystem service category, we tested whether a more stringent classification would alter the results of the second-order meta-analysis. We therefore reclassified the response variables into 17 ecosystem service categories, keeping separated ecosystem functions, physical properties, variables related to organisms, above- and belowground variables, etc. For example, soil fertility was divided into soil physical characteristics, soil nutrient availability and soil organisms, biodiversity into above- and belowground biodiversity, water regulation into water quality and water quantity; climate regulation was divided into variables related to emissions and uptakes, and crop yield into crop yield and crop biomass. Results were robust as shown by similar effect sizes compared to broader classification (fig. S6). The only deviation from previous results is represented by crop biomass, which was significantly increased under diversification regimes. The eight effect sizes included in this category referred to both above- and belowground biomass. This sensitivity analysis confirms that organisms both above- and belowground strongly respond to diversification practices. The exclusion of single or multiple diversification practices and single or multiple ecosystem service categories did not affect our results (results not presented).

### SUPPLEMENTARY MATERIALS

Supplementary material for this article is available at <http://advances.sciencemag.org/cgi/content/full/6/45/eaba1715/DC1>



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## Agricultural diversification promotes multiple ecosystem services without compromising yield

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